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EDITORIAL

BOVINE MASTITIS

With the introduction of new methods of preventing and controlling the different types of bovine mastitis, following upon the study of the use of new chemical compounds and antibiotics, there may be a tendency to give less attention to dairy and cowshed hygiene in controlling the various associated infections. It is opportune, therefore, to draw attention to the article published in this issue of the British Veterinary Journal, which describes experiments designed to show the value of a series of agents in destroying *Streptococcus agalactiæ*, one of the micro-organisms commonly found in the udder and on materials soiled by infected milk. In studies on bovine mastitis it becomes obvious that a cause of "breakdown" in a herd freed from infection, so far as the udder is concerned, is the re-introduction of the infection from an outside source. Such sources are many and varied: they include the skin of the cow, milkers' hands and wearing apparel, as well as objects in the cowsheds. The so-called "blitz" treatment, which is now adopted as part of the treatment of the disease, includes a thorough disinfection of all such objects. Such treatment, well carried out, has been shown to be of considerable value in preventing re-infection of the udder, and is to be recommended as a routine measure in controlling the infection. The choice of a satisfactory agent at an appropriate dilution merits attention. It may not be of any great importance to select a special agent for the disinfection of the cowshed and objects within the cowshed, provided an adequately good "strength" is used; but for skin disinfection much care is necessary in the selection in order that the agent will destroy the micro-organisms when used in a dilution which will produce no harmful effects on the skin or other tissues with which it is brought into contact. The results now published are, therefore, valuable contributions to our knowledge of the subject.

Another point to which attention may be directed concerns the persistence of *Str. agalactiæ* in sores on teats. It is now becoming clear that such sores

may continue to harbour *Str. agalactiæ* for long periods, and are responsible for persistent or intermittent infection of the udder. Failure of a quarter or an udder to respond to the now generally adopted treatment with penicillin or other antibiotics by injection through the teat canal may, of course, be due to a variety of causes. The presence of teat sores, however, should always be taken into consideration in dealing with such failures: it is a wise policy in practice to treat them by the application of appropriate agents.

The control of bovine mastitis has now reached a stage when it becomes a practical and economic proposition. It is to be expected that completely successful results may not be the outcome of any method of treatment, but the results of experiments and experiences in the field are sufficiently good to show that *Str. agalactiæ* infection in the udder can be greatly reduced even in a heavily infected herd and can be maintained at a low incidence. Although equally satisfactory results cannot be reported yet in dealing with other infections, further research work with the ever-increasing list of antibiotics will, we hope, lead to the position when routine treatments can be applied with equally good effects. It is freely acknowledged and recognised that bovine mastitis in its various forms is now one of the most important limiting factors governing maximum milk production in the dairy industry. Without doubt, in time and, we believe, in a short time, bovine mastitis, in all its varieties, will be added to the list of diseases capable of being well controlled and for which practical methods of eradication have been worked out.

GENERAL ARTICLES

THE BACTERICIDAL EFFECT OF VARIOUS DISINFECTANTS ON *STR. AGALACTIÆ* ON THE SKIN AND IN THE ENVIRONMENT OF THE COW

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THE eradication of *Str. agalactiæ* from infected herds often presents difficulties, in spite of effective treatment of individual animals, largely because of the frequency and persistence of the organisms outside the bovine udder.

The survival time of *Str. agalactiæ* on various objects and materials has been shown by many workers to be from a few days to several months: Bryan (1934), Harrison (1941), Klingmuller (1930), Report 2, Imperial Bureau of Animal Health (1944), Watts (1944). Munch-Peterson, *et al.* (1940), found large numbers of *Str. agalactiæ* in sores on teats and udder. Chodkowski (1949, in the press) has demonstrated that *Str. agalactiæ* can survive on parts of the skin and on various objects in an infected cowshed, for periods up to three weeks.

Taking the work of Price (1938) as an indication, it seems likely that *Str. agalactiæ* is present only temporarily unless it multiplies in sites such as sores on the teats, when it may continue to survive. In the work here reported consideration was given to skin autodisinfection, i.e., spontaneous disappearance of the organism from the skin. In this respect Norton and Novy (1931) and Arnold, *et al.* (1930), showed that a proportion of the organisms implanted on to the human skin disappear without the use of any disinfectant. If the skin or some part of it which is more liable to outside soiling is covered with all kinds of dust, dirt, grease, etc., various pathogenic and nonpathogenic bacterial strains will survive there for a longer period. After removing the dirt and the grease from the skin, by using soap and water or any detergent, the power of the outer layer of the skin destroys the temporary pathogenic and nonpathogenic bacterial flora within a short period of time. Because of the well-recognised fact that the action of disinfectants should be tested in the presence of organic matter, this was given due consideration in the present work; *Str. agalactiæ* outside the udder is almost always attached to organic matter of various kinds, e.g., protein, grease, hay, bedding, dust, etc., which protects the organism, and so the power of disinfecting agents is reduced.

According to Colebrook (1941), transient flora, when implanted on the skin, disappears spontaneously, or can be removed by washing and antiseptics. Baker (1941) describes the bactericidal action of synthetic detergents on gram-positive and gram-negative organisms *in vitro*. Williams (1943), gives the results of the antiseptic effect of CTAB on human skin and wounds and the sterilising effect on surgical instruments and utensils. Barnes (1942) reduced the number

CTAB, or "Cetavlon," is the trade mark for a quarternary ammonium compound and a synthetic cationic detergent, containing 75 per cent of cetyl-trimethyl-ammonium bromide.

of bacteria on normal skin and was able to sterilise dirty bowls and baths by using CTAB solution.

The purpose of this work was to find the most effective disinfectant, and the concentration, base and mode of application, which would destroy *Str. agalactiæ* outside the udder, encourage the healing of *Str. agalactiæ* infected sores on the teats and be harmless to the skin and mucous membrane of the eye.

Many experiments were carried out, but only the most representative will be described.

Media

Four media were employed: (a) Sterile milk. (b) Udder broth, prepared (i) in a manner similar to ordinary meat extract broth, except that instead of meat, lactating or non-lactating udder tissue was employed; (ii) as a trypsin-digest udder broth, made according to modified Douglas trypsin broth. (c) Two selective media (Chodkowski and Lancaster, 1949), viz.: (i) a selective milk medium, consisting of sterile milk containing potassium tellurite 1 : 20,000 and boracic acid 1 : 500, final concentrations; and (ii) selective solid medium, consisting of sheep blood-agar plates, containing crystal violet 1 : 750,000, thallium sulphate 1 : 3,000, and æsculin in 1 : 1,000, final concentrations.

Methods

(a) *In vitro.* A number of tubes, containing one or other of the above media, were inoculated with known numbers of *Str. agalactiæ.* After incubation for two to six hours at 37 degrees C., a loopful of the contents of each tube was inoculated on a blood agar plate to test for the presence of the organism. Measured amounts of different disinfectants were added to each of the tubes, and the contents were then thoroughly mixed and incubated at 37 degrees C. At various intervals of time thereafter a small quantity of the mixture from each tube was plated on blood agar in order to test the survival of the organisms. The plates were read after incubation at 37 degrees C. for 48 hours and the colonies identified. In these experiments milk medium was included, because *Str. agalactiæ* is spread naturally by infected milk, so contaminating the environment, and because the effects of disinfectants on organisms in the presence of the organic matter in milk is in some respects similar to the condition in which these disinfectants act on the skin. In the first tests, sodium oleate was used in adequate proportion in an attempt to neutralise the action of CTAB on the organisms in liquid milk media. Tests were carried out in two series of tubes containing *Str. agalactiæ* milk cultures and CTAB in varying concentrations. Sodium oleate was added to one series. Both series of tubes failed to show any difference in viable count, and so the use of sodium oleate was discontinued in subsequent experiments.

(b) *In vivo.* The teats and shaved and unshaved areas of the skin were used. The skin of the upper parts of the left and right flanks was shaved to form rectangular areas parallel to the spine 4″ by 21″ in area. This was done at least two days before the commencement of the experiment. All the areas were examined for the presence of *Str. agalactiæ,* according to the method

described by Chodkowski and Lancaster (1949). The selected areas were then rubbed with cotton-wool swabs moistened in *Str. agalactiæ*-naturally infected milk, or in 24 hours serum broth culture of *Str. agalactiæ*, once a day for three consecutive days in order to implant the organisms on the skin. On the third day following the last implantation, the areas were left to dry for about 15 minutes and were then swabbed to prove the presence of implanted organisms. The shaved areas were divided into squares of four inches and marked with grease pencil, for identification. An estimated amount of a known concentration of different disinfectants was then rubbed on each of these areas by means of cotton-wool swabs. Tests for the duration of survival of *Str. agalactiæ* were carried out at various intervals, using the previously described swabbing technique, until three negative results were obtained. Since the disinfectants and the previously applied soap, in addition to natural skin autodisinfection, have an inhibitory effect on the organisms on the skin, sterile milk without dyes was used to eliminate further inhibitory action.

The Effect of Various Disinfectants on Str. Agalactiæ in Vitro

Four series of tubes of different liquid media were used. Each of three series, i.e., sterile milk, selective milk medium and udder broth were inoculated with 0.1 ml. of *Str. agalactiæ* broth culture, containing approximately two million organisms, and were incubated for four hours at 37 degrees C. along with a fourth series of tubes containing *Str. agalactiæ*-naturally infected milk. Tests were carried out as described under "Technique *in vitro*."

TABLE I.

Time of Action of Disinfectant		15 mins	2½ hrs.	18 hrs.	2½ hrs.	18 hrs.	2½ hrs.	18 hrs.	18 hrs.
Disinfectant	Final Concent	Milk			Selective Liquid Medium		Udder Broth		Str. agal. natual infected milk
Acriflavine	1:25,000	0	+++	+++	+++	+++	+++	+++	+++
Chloros	1:50	+++	++	++	++	++	-	-	+++
CTAB	1:500	-	-	-	-	-	-	-	-
CTAB	1:1000	-	-	-	-	-	-	-	-
Dettol	1:500	0	+++	+++	+++	+++	-	-	+++
Iodine 3% in 70% Spirit	1:500	0	-	-	-	-	-	-	-
Iodine 3% in 6% K.I. aqueous solution	1:800	0	+	-	-	-	-	-	0
Iodine 4% in ointment	1:600	0	+++	+++	+++	++	+++	+++	+++
Liquid Soap	1:25	0	+++	++	+++	+++	-	-	+++
Methylated Spirit	1:25	0	+++	+++	+++	+++	+++	+++	+++
Sulphapyridine	1:65	0	+++	++	+++	+++	+++	+++	+++
Penicillin	100 units per ml.	0	+	+	+	-	+	-	-
Teepol	1:500	+++	+++	+++	0	0	0	0	0
Teepol and Chloros	1:500, 1:50	+++	+++	+++	0	0	0	0	0
Lysol	1:100	0	0	++	0	0	0	0	0

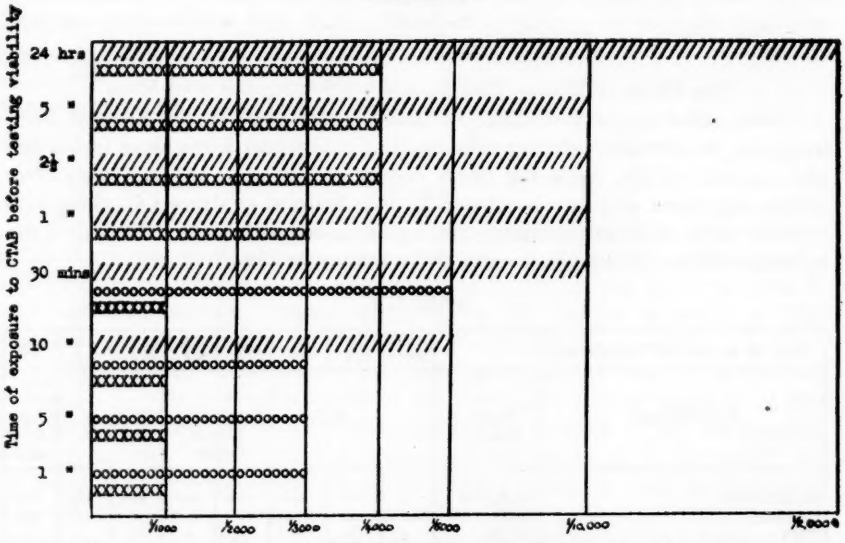
O = Not tested.
— = No growth.
\+ = 1-20 colonies of *Str. agalactiæ*.
++ = 20-100 colonies of *Str. agalactiæ*.
+++ = 100-500 colonies of *Str. agalactiæ*.

Table I shows that the most effective disinfectants for *Str. agalactiæ* in milk and in broth culture are CTAB, 1 : 500 to 1 : 1,000, final concentrations, and iodine solution 1 : 500 to 1 : 800, final concentrations. The other disinfectants appeared to be less effective, more especially in milk than in broth cultures.

The Effect of CTAB on Str. Agalactiæ in Vitro

The effect of CTAB on *Str. agalactiæ* in whole milk, in milk with 90 per cent plain water and in udder broth was studied. Each of the series of tubes containing the three different media was inoculated with 0.2 ml. of 24 hours' serum broth culture of *Str. agalactiæ* incubated for two hours at 37 degrees C.: the test was carried out as in the previous experiment.

TABLE II.



Various concentrations of CTAB lethal to Str. agalactiæ after exposure at 37 degrees C. for various lengths of time.

Concentration of CTAB

XXXXXX represents milk culture.
oooooo " 10 per cent milk culture in water.
////// " broth culture.

Each horizontal column represents those concentrations of CTAB which were lethal to *Str. agalactiæ* within the stated time.

Table II shows that the concentration of CTAB lethal for *Str. agalactiæ* was 1/1,000 in whole milk within one minute, 1/3,000 within one hour, and 1/4,000, final concentration, from 2½ hours upwards. The lethal concentration of CTAB for *Str. agalactiæ* in 10 per cent milk culture in water was 1/3,000 in one minute and 1/5,000 within 30 minutes. The bactericidal concentration of CTAB for *Str. agalactiæ* in broth was 1/5,000 within 10 minutes, 1/10,000 within 30 minutes, and 1/20,000 within 24 hours.

The Effect of Disinfectants on Str. Agalactiæ-infected Skin

The teats and shaved flanks of an eight-year-old cow, Lo., were infected with 24 hours' *Str. agalactiæ* serum broth cultures once a day for three days. After disinfection, swabs were taken at intervals and agitated in the milk media and udder broth, these media being examined as before.

TABLE III.

Area	Disinfectants	Amount Applied on the area	15 mins.	2 hrs.	45 mins.	5½ hrs.		22½ hrs.
			Milk	Milk	Udder Broth	Milk	Udder Broth	Milk
Teat LF	- Control	-	++	++	+++	+	++	+
" LH	Penicillin Cream	450 mgms	+	+	+	+	+	+
" RF	1% CTAB aqu. sol.	2 mls	-	-	-	-	-	-
" RH	3% Iodine in 70% spirit	2 "	-	-	-	-	-	-
4" square flank areas A	- Control	-	++	+++	+++	++	++	++
B	4% Iodine ointment	400 mgms	+	+	+	+++	++	++
C	0.1% Acriflavine	2 mls	++	++	++	++	++	++
D	20% Sulphapyridine	1 ml.	++	++	+++	+++	++	++
E	5% Dettol	2 mls	++	++	+	+	++	+
F	1% Liquid Soap	2 "	++	++	+	+	++	+
G	Meth. Spirit	2 "	+	+	+	+	++	+
H	3% Iodine in 6% K.I. aqueous solution	2 "	-	-	-	-	-	-
I	2% Chloros	2 "	+	++	+	+	++	++
J	Water	2 "	++	++	++	++	++	++
K	- Control	-	+++	+++	+++	+++	+++	+++

— = No growth.
\+ = 1-20 colonies of *Str. agalactiæ*.
++ = 20-100 colonies of *Str. agalactiæ*.
+++ = 100-500 colonies of *Str. agalactiæ*.

Table III shows that 1 per cent CTAB aqueous solution, 3 per cent iodine in 70 per cent spirit, and 3 per cent iodine in 6 per cent potassium iodide aqeuous solution sterilised the areas in 15 minutes, while the other disinfectants, e.g., penicillin cream (1,000 units per gram), 4 per cent iodine ointment, 0.1 per cent acriflavine, 20 per cent sulphapyradine, 5 per cent Dettol, 2 per cent Chloros* (2,000 p.p.m. of available chlorine) methylated spirit and 1 per cent liquid soap did not appear to have much bactericidal effect on *Str. agalactiæ* on the skin, since the organisms were recovered up to 22 hours after the disinfectants had been applied.

The Effect of CTAB, Soap, Teepol and Chloros on the Skin

In this experiment the same cow as in Experiment 3 was used. Flank areas only were infected on three consecutive days with *Str. agalactiæ*-naturally infected milk—and the test was carried out as in Experiment 3. The results showed that 1 and 0.5 per cent aqueous solution of CTAB alone, or mixed with

* Chloros is the trade mark for sodium hypochlorite, containing about 10 per cent of W/W available chlorine up to three months from the date of despatch, when stored in a dark, cool place. All Chloros used in these tests was stored in this way.

1 per cent soap or with 0.2 per cent Teepol,* or with 2 per cent Chloros, killed *Str. agalactiæ* on the skin within 15 minutes, while soap, Teepol and Chloros used separately and a mixture of Chloros and Teepol of the above strength used without CTAB failed to kill the organisms within 24 hours. The control, untreated areas yielded *Str. agalactiæ* up to 24 hours.

The Effect of Aqueous Solution of CTAB, CTAB Cream, Penicillin Cream and Iodine on the Skin

In this experiment two different animals were used, a six-year-old cow, G.1, and a three-year-old heifer, G.2. The flanks of both animals were shaved and infected with *Str. agalactiæ*-naturally infected milk, except one flank of heifer G.2, which was infected with a 24 hours' serum broth culture of *Str. agalactiæ*. The experiment was then carried out as in the previous tests. It was found that 1, 5 and 10 per cent aqueous solution of CTAB, 5 per cent CTAB

TABLE IV.

Infected With				Serum and Glucose Broth Culture					Str.agalactiae naturally infected					Serum Broth Culture			
Animal				Cow Lo. 32					Cow G.1					Heifer G.2			
Area			Disinfectant	Swabbed after					Swabbed after					Swabbed after			
				1½ hrs	24 hrs	48 hrs	72 hrs	96 hrs	1½ hrs	24 hrs	48 hrs	72 hrs	96 hrs	1½ hrs	24 hrs	48 hrs	72 hrs.
Left Flank		A	0.05% CTAB aqueous solution	+	+	+	+	+	+	+	+	-	-	++	+++	-	-
"	"	B	0.1% " " "	+	+	+	+	-	+	+	+	+	-	+++	++	+	-
"	"	C	0.2% " " "	+	+	-	-	-	+	+	+	+	-	+	+	-	-
"	"	D	0.5% " " "	+	+	-	-	-	+	+	-	-	-	+	+	-	-
"	"	E	1% " " "	-	-	-	-	-	-	-	-	-	-	-	-	-	-
"	"	F	2% " " "	-	-	-	-	-	-	-	-	-	-	-	-	-	-
"	"	G	5% " " "	-	-	-	-	-	-	-	-	-	-	-	-	-	-
"	"	H	10% " " "	-	-	-	-	-	-	-	-	-	-	-	-	-	-
"	"	I	5% Chloros	+++	+++	+++	+++	+++	++	++	-	-	-	+	+	+	+
"	"	J	- Control -	+++	+++	+++	+++	+++	+++	+++	++	++	-	++	++	++	+
Right	"	K	4% Iodine Ointment	+	-	-	-	-	+++	+++	+	+	++	+++	++	-	-
"	"	L	Penicillin Cream 1000 units/grm.	+	-	-	-	-	-	-	-	-	-	0	0	0	0
"	"	M	0.05% CTAB Cream	+	+	+	+	-	+	+	+	+	-	+	+	+	+
"	"	N	0.1% CTAB Cream	+	+	+	+	+	+	+	+	+	-	+	+	+	-
"	"	O	0.2% " "	+	+	+	+	-	+	+	+	+	+	+	+	-	-
"	"	P	0.5% " "	+	+	-	-	-	+	+	+	+	-	+	+	-	-
"	"	Q	1% " "	+	+	-	-	-	+	+	+	+	+	+	+	+	+
"	"	R	2% " "	+	+	-	-	-	+	+	+	-	-	+	+	-	-
"	"	S	5% " "	-	-	-	-	-	-	-	-	-	-	-	-	-	-
"	"	T	10% " "	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Teats	L.F.		10% " "	-	-	-	-	-	-	-	-	-	-	0	0	0	0
"	L.H.		Penicillin Cream 100 units/grm	+	+	-	-	-	-	-	-	-	-	0	0	0	0
"	R.F		- Control -	+	+	-	-	-	+	-	-	-	-	0	0	0	0
"	R.H		2% CTAB Cream	+	-	-	-	-	+	+	-	-	-	0	0	0	0

\+ = 1-20 colonies of *Str. agalactiæ*.
++ = 20-100 colonies of *Str. agalactiæ*.
+++ = 100-500 colonies of *Str. agalactiæ*.
++++ = Above 500 colonies of *Str. agalactiæ*.
— = No growth.
O = Not tested.

* Teepol is a trade mark for a synthetic cleaning detergent.

cream in lanoline, lanette wax and paraffin base, penicillin cream (1,000 units per gram) and 3 per cent iodine in 70 per cent methylated spirit, destroyed *Str. agalactiæ* on the skin within 2½ hours, while the other disinfectants used, failed to kill these organisms within 72 hours. The control, non-treated areas on both cows yielded *Str. agalactiæ* up to 72 hours.

Estimation of the Minimum Bactericidal Concentration of CTAB on Str. Agalactiæ-infected Skin

In order to estimate the most effective concentration of CTAB in aqueous solution and in cream on the shaved skin of the flank, and on the skin of the teats, three animals were used. The teats and shaved flanks of cow Lo. and the shaved flanks of heifer G.2 were infected with 24 hours' *Str. agalactiæ* serum broth culture, and the cow G.1 with *Str. agalactiæ*-naturally infected milk. Tests were carried out as in previous experiments.

As seen from Table IV, the efficiency of CTAB concentrations in aqueous solutions and in cream, whether used on the flanks or on the teats, appeared to be similar on the skin of all three animals. The lowest effective concentration of CTAB aqueous solution appeared to be 1 per cent. The maximum concentration of CTAB cream which failed to sterilise the skin was 2 per cent, and the minimum tested concentration which killed *Str. agalactiæ* on the skin was 5 per cent. The higher concentration of CTAB cream (between 1 and 5 per cent) necessary to sterilise the *Str. agalactiæ*-infected skin as compared with CTAB aqueous solution (1 per cent) was probably caused by the presence of lanette wax, a dispersing agent in the base of the cream which partly inhibits the action of CTAB (Slavin, 1949). Penicillin cream (1,000 units per gram) appeared to be less effective; 4 per cent iodine ointment and 5 per cent Chloros aqueous solution showed very little bactericidal effect on *Str. agalactiæ* on the skin.

Comparison of the Effect of Spraying and Brushing CTAB and Chloros on Str. Agalactiæ-infected skin, and the Effect of Cresol on the Loose-box*

(a) Selected areas of the skin of cow G.1 and heifer G.2, especially the upper part of the tail, base of horns, the flanks, legs and teats and the loose-box were infected with *Str. agalactiæ*-naturally infected milk—on three consecutive days—by the use of large cotton-wool pads, to implant the organisms on to the skin and hair. On the last day of infection the presence of the organisms was confirmed, and the whole of the skin of cow G.1 was sprayed thoroughly with 0.5 per cent CTAB aqueous solution and the skin of heifer G.2 with 2 per cent Chloros. After transferring the animals to a field, the loose-box was sprayed with 5 per cent cresol, and the disinfected animals were then returned to it. On the following day, swabs were taken from the various areas of the skin and from the loose-box to test for the presence of *Str. agalactiæ*. Swabs from the cow G.1 were negative, while the swabs from heifer G.2 were positive, *Str. agalactiæ* being recovered from three out of the seven swabs, viz., from the base

* "Loose-box" includes manger, cleaning utensils, buckets, milking stools, etc.

of the horns, base of the tail and from the neck, where the organisms survived for two days. *Str. agalactiæ* was also recovered up to the fifth day from the window ledge of the loose-box, in spite of spraying with 5 per cent cresol.

(b) The same animals were used as in (a), but at a later time. After infection, both animals were disinfected by spraying and brushing, particular attention being paid to the previously infected areas. Cow G.1 was treated with 2 per cent Chloros and heifer G.2 with 0.5 per cent CTAB aqueous solution. Swabs taken from the skin of both animals on three consecutive days were negative. It appears, therefore, in comparison with (a), that efficient and careful spraying and brushing, with particular attention to infected areas, gave better results than spraying alone. Even Chloros, which is less efficient than CTAB, killed *Str. agalactiæ* on the skin by this method.

(c) Selected parts of the skin of four cows were infected with *Str. agalactiæ*-naturally infected milk. Cow Lo. was sprayed with 0.5 per cent aqueous solution of CTAB, and 1 per cent CTAB solution was brushed on to the skin of cow 928. Cows 964 and 968 were sprayed with 1 per cent CTAB solution. Swabs taken after treatment from cows Lo. and 928 were negative for three consecutive days. *Str. agalactiæ* could not be recovered from cows 964 and 968 at 24 hours, and when they were artificially reinfected, 24 hours after disinfection, no *Str. agalactiæ* was recovered on three consecutive days. This shows that 0.5 per cent CTAB aqueous solution was efficient and bactericidal for *Str. agalactiæ,* and that the disinfecting power of CTAB on the skin lasted for at least 24 hours.

(d) The skin of cow Lo., and the loose-box were infected on three consecutive days with *Str. agalactiæ*-naturally infected milk. After swabbing all areas to confirm the presence of *Str. agalactiæ,* the cow was sprayed with 0.5 per cent CTAB and the loose-box with 0.2 per cent CTAB aqueous solution. Swabs taken on three consecutive days were negative for *Str. agalactiæ,* except those from the hairy base of the horn, where the organisms survived up to 24 hours, but were absent at 48 hours. This suggests that some areas of skin, e.g., massed hair at the base of the horn, neck, upper part of the tail, are more difficult to disinfect, and require special attention as compared with the udder and teats, which are comparatively easy to disinfect.

(e) Twenty-four cows in the Laboratory cow-shed were used for this experiment. The base of their horns, upper parts of the tails and the teats of each cow were heavily infected with *Str. agalactiæ*-naturally infected milk on three consecutive days. Twelve of these cows were disinfected with 1 per cent CTAB aqueous solution, six of which were sprayed and six brushed. The other twelve cows were disinfected with 2 per cent Chloros (2,000 p.p.m. available chlorine), six being sprayed and six brushed. The result of this experiment showed that Chloros failed to remove *Str. agalactiæ* completely on three out of twelve cows, one brushed and two sprayed. The organisms survived on the base of the tails of two cows up to 24 hours, and at the base of the horns of one cow, up to two days. All twelve cows sprayed and brushed with CTAB solution were negative,

with the exception of cow 988, in which an unhealed sore on the teat did not respond to penicillin cream, nor to 3 per cent CTAB cream treatment.

The Effect of Iodine Vapour on Str. Agalactiæ-infected Skin and Loose-box

The experiment consisted of two consecutive tests, in each of which the same cow and loose-box were used to test the effect of iodine vapour. *Test* (*A*): The cow and loose-box were infected with *Str. agalactiæ*-naturally infected milk —once a day on three consecutive days. Daily examinations were carried out until three negative tests had been obtained, to estimate the duration of survival of the organisms. *Test* (*B*): The same cow and loose-box were again infected with *Str. agalactiæ* on three consecutive days. Six hundred g. iodised carbon (96 per cent iodine) was exposed in six Petri dishes in various positions, including suspension from the ceiling. The loose-box was kept closed, except for the opening of the upper part of the door which was necessary for ventilation, and the test was carried out as previously described.

The results of these tests showed that *Str. agalactiæ* survived under natural conditions on the body up to eight days (test A), and when exposed to iodine vapour up to seven days (test B). The amount of iodine in the air of a loose-box after one week's exposure of iodine carbon was 6.5 μg. per litre, and 20 mg. of iodine was found per 100 grams of cow's hair. In 100 grams of hair from a normal cow housed in the next loose-box, less than 0.1 mg. of iodine was found.

The Bactericidal Effect of Disinfectants on a Str. Agalactiæ-contaminated Loose-box and on Dairy Utensils, Milkers' Clothing, Udder Cloths, and Teat Cups, etc.

(a) The effect of formaldehyde on *Str. agalactiæ* was tested. Materials and equipment usually found in a cowshed and dairy, e.g., cleaning utensils, milking utensils, milkers' clothing, woollen and rubber clothes, boots, paper, sacks, cow's hair, straw, hay, dung, bricks, concrete, etc., were placed in a sealed room of 342 cu. ft. capacity. The interior of the room, including the lime-washed wall and the contents, were smeared with *Str. agalactiæ*-infected milk on three consecutive days. A glass dish containing 225 gr. potassium permanganate was placed on the floor of the room, and into this was poured 225 ml. formalin. The room was then quickly sealed and the contents left exposed overnight to the penetration of formaldehyde so produced. *Str. agalactiæ* was not recovered from the swabs taken from the different contents of the room on the following day. It seems, therefore, that formaldehyde is highly effective as a bactericidal agent against *Str. agalactiæ*.

(b) The effect of disinfectants on *Str. agalactiæ*-contaminated udder cloth was tested. A number of pieces of udder cloth, $\frac{1}{2}''$ x $\frac{1}{2}''$, were sterilised by autoclaving, dried, and soaked for one hour in *Str. agalactiæ*-infected milk. Each piece was treated with a different disinfectant for a different period of time, then immersed in sterile milk medium and incubated at 37 degrees C., following which samples were plated on solid medium. *Str. agalactiæ* was destroyed on the udder cloth within five minutes when treated with 1 per cent CTAB aqueous

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solution, within 30 minutes when treated with 0.5 per cent CTAB solution, and within one hour when treated with 0.1 and 0.2 per cent CTAB solutions. Up to five hours' treatment with 2 per cent Chloros (2,000 p.p.m available chlorine) was necessary to destroy *Str. agalactiæ* on the udder cloth. *Str. agalactiæ* were still present on the udder cloth after 48 hours' treatment with tap water only or with 2 per cent solution of liquid soap.

(c)) The effect of disinfectants on *Str. agalactiæ*-contaminated milk bottles was tested. Each of seven sterile bottles was subjected to contact with *Str. agalactiæ*-infected milk for one hour. After removing the milk, each bottle was rinsed for one minute with a different disinfectant. After pouring off the disinfectants, each bottle was rinsed with 20 ml. sterile milk, which was transferred to a McCartney bottle, incubated at 37 degrees C., and then plated. The organisms were destroyed within one minute when washed with 1, 0.5, 0.2 and 0.1 per cent aqueous solution of CTAB, but they were recovered from bottles when treated in a similar manner with plain water, 2 per cent soap, or 2 per cent Chloros.

(d) The effect of disinfectants on *Str. agalactiæ*-contaminated milking buckets was tested. Seven sterilised buckets were contaminated with *Str. agalactiæ*-infected milk for 30 minutes. After removing the milk, the buckets were rinsed with plain water for one minute and then each was rinsed with a different disinfectant for 30 seconds. After pouring off the disinfectants, each bucket was rinsed with sterile milk, which was transferred to glass jars, incubated at 37 degrees C., and plated. The organisms were destroyed when the buckets were treated with aqueous solution of 1.0 and 0.1 per cent CTAB, or with 70 per cent methylated spirit, but they were recovered following treatment with plain water only, 2 per cent soap, or 2 per cent Chloros.

(e) The effect of disinfectants on *Str. agalactiæ*-contaminated hand brushes was tested. Eight sterilised brushes were contaminated with *Str. agalactiæ*-infected milk for 30 minutes. The brushes were rinsed in plain water for three-five seconds and then each was rinsed in a different disinfectant. Following this, each brush was dipped three times in sterile milk medium which was incubated at 37 degrees C. and plated. *Str. agalactiæ* was destroyed on the hand brush treated with 1.0 and 0.1 per cent CTAB aqueous solution: the organism was obtained from the brushes treated in a similar manner with plain water only, 70 per cent methylated spirit, 2 per cent soap, or 2 per cent Chloros.

(f) The effect of disinfectants on *Str. agalactiæ*-contaminated milking machine teat-cups was tested. Eight teat-cups, sterilised by steam in the usual manner in the dairy, were placed in *Str. agalactiæ*-infected milk for one hour. Each contaminated cup was dipped three times in plain water and then three times in a disinfectant. The cups were washed in sterile milk medium, which was then incubated at 37 degrees C. and plated. The organisms were destroyed within three seconds by 1, 0.5 and 0.1 per cent CTAB aqueous solution, but they were recovered from the teat-cups, treated in similar manner with plain water only, 70 per cent methylated spirit, or 2 per cent Chloros.

The Effect of CTAB on Cows' Skin and the Mucous Membrane of the Eye

CTAB was used as a solution in saline or tap water on the mucous membrane of the eye and on the cows' skin, and as a cream with lanoline, lanette wax and paraffin base, on the skin of the teats, udder and flanks.

(a) The Effect of CTAB on the Mucous Membrane of the Eye

Various concentrations of CTAB solution in saline or in tap water were instilled in 0.5 to 1 ml. quantities with the aid of a syringe under the lower lid of one or both eyes. Occasionally, for control purposes, one eye was instilled with CTAB solution and the other eye with saline only.

Reactions were observed for 48 hours and the results recorded. A test was carried out on the mucous membrane of the eyes of four animals, three cows and one heifer, commencing with a 0.001 per cent solution of CTAB in saline and gradually increasing to a 5 per cent solution in water used during a period of several days. No reaction was observed on the mucous membrane of these animals from 1½ up to 24 hours when using concentrations of CTAB up to 3 per cent. A definite reaction was observed from 1½ up to 24 hours after treatment when using 4 and 5 per cent CTAB solutions, but the symptoms disappeared within 48 hours without any permanent injury to the eye. Finally, a test was carried out on 24 cows at the laboratory using 0.5, 1 and 2 per cent CTAB aqueous solution in order to find the maximum concentration which would be harmless. A slight transitory irritation to the mucous membrane of the eye of some cows was found with 1 per cent within 30 minutes of application, and the symptoms disappeared in less than two to six hours; while using 2 per cent a slight reaction started within 30 minutes from the beginning of the treatment and lasted for less than 24 hours. One cow (1198) was particularly sensitive to 2 per cent CTAB solution, but even here the symptoms disappeared within 24 hours without lesions or apparent ill effect on the eye.

(b) The Effect of CTAB on the Skin of the Teats

Various concentrations from 0.02 to 10 per cent CTAB in aqueous solution or in cream were applied once or twice daily with the aid of cotton-wool swabs on the skin of the teats of four cows, for periods of five, seven and eight days. The reaction of the skin was observed, and the results were recorded during and for two days following the final treatment. CTAB aqueous solution of the above concentrations did not cause any visible lesion on the skin of the teats, except that after seven days' treatment with 5 or 10 per cent CTAB solution, the skin appeared to become dry and glossy. These symptoms disappeared in one or two days after discontinuing the use of the disinfectant. One to 10 per cent CTAB cream applied daily on the teats for seven or eight consecutive days did not have any apparent ill effect either during treatment or up to three days after treatment.

(c) The Effect of CTAB on the Skin of the Udder

Two cows were used to test the effect on the skin of the udder of CTAB aqueous solution and CTAB cream. On cow Cn, 1, 2, 5 and 10 per cent CTAB

aqueous solutions were applied to the four quarters of the udder skin on four consecutive days. With 2 per cent CTAB solution, slight reaction (redness, heat) appeared after 48 hours and lasted for two days after the final application. Stronger reaction (redness, pain and swelling) appeared after 24 hours when 5 and 10 per cent were used, and the reaction was present up to the last day of observation, i.e., two days after the final application. The use of 1 per cent CTAB aqueous solution on four consecutive days did not have any apparent ill effect, although the udder appeared to be more sensitive to these solutions than did other parts of the cow's body. On cow Lo., 2, 3, 4, 5 and 10 per cent CTAB cream was applied to various parts of the skin of the udder, once a day for three consecutive days. With 2 per cent a slight reaction appeared on the fourth day; with 3 per cent a stronger reaction (redness, pain) appeared on the fourth day; with 4 per cent and with 5 per cent a strong reaction (redness, heat and pain) appeared on the third day; and a very strong reaction (redness, heat, pain and swelling) appeared on the third day when 10 per cent was applied. These reactions persisted up to two days after the last application; observations then ceased.

(d) The Effect of CTAB on the Skin of the Flank

Areas of shaved skin on the flanks of four cows were used for this experiment, and the effect of CTAB in aqueous solution and in cream was observed.

(1) One, 2, 3, 4, 5 and 10 per cent CTAB aqueous solutions were applied to the skin once a day for three to seven consecutive days. Three per cent solution caused a reaction (dark discoloration and pain) on the seventh day, while 4 and 5 per cent solutions caused a similar reaction on the second and third day after the commencement of the treatment. A strong reaction (swelling, dark discoloration, pain and heat) was observed when 10 per cent was used. One cow and a heifer did not react to the 5 per cent solution, and the heifer showed only slight reaction on the fourth day following application of 10 per cent CTAB solution. When 1 and 2 per cent CTAB solutions were used no apparent ill effect was caused to the skin.

(2) When 5 and 10 per cent CTAB cream was applied to the skin of the flank of four cows for three consecutive days, one cow reacted very markedly (heat, dark discoloration, swelling, pain) to 10 per cent CTAB cream after three days, but the reaction was less when 5 per cent was used. The second cow showed no reaction to 5 and 10 per cent after three days. The third cow showed no reaction to 5 per cent, but after three daily applications of 10 per cent CTAB cream a definite reaction was observed. The skin of the flank of the fourth animal, a heifer, failed to show any reaction during three daily applications of 5 and 10 per cent CTAB cream and during the following four days.

It appears, therefore, from the above experiments that 1 per cent CTAB aqueous solution can be safely used for the disinfection of the skin; this concentration has also been shown to be harmless to the mucous membrane of the eyes. Treatment of the skin of the teats, flank and udder with 3 per cent, 2

per cent and 1 per cent CTAB cream, respectively, can be continued up to three or four weeks, providing observations are made during the treatment, since some cows may show particular sensitivity to CTAB. In such sensitive animals, treatment should be discontinued whenever signs of irritation are seen.

Use of Disinfectants in the Treatment of the Sores on Cows' Teats

(a) Positive Milk Samples Due to a Str. Agalactiæ-infected Teat Sore; It's Cure with Penicillin Cream, Followed by Negative Samples

Milk samples from the cow concerned had consistently shown *Str. agalactiæ*. Within a period of 11 days the cow received two courses of intramammary infusions, each of five daily injections of 100,000 units of penicillin, and during this period the teats were treated with penicillin cream containing 1,000 units per gram. In spite of this treatment, milk samples continued to show infection. During the period of treatment it was observed that the cow had, at the tip of the left hind teat, a wet and deeply ulcerating sore which penetrated the teat canal. This sore was found to be regularly infected with *Str. agalactiæ*, and impression cultures from it consistently showed large numbers of these organisms. Milk samples from this quarter also regularly showed heavy *Str. agalactiæ* infection, while samples from other quarters were negative. It seemed possible that the samples from the left hind quarter were becoming contaminated during the passage of the milk over the infected sore. By inserting a large syphon into the teat and then sampling by means of a small syphon passed through the larger one, it was shown that contamination of the milk sample was occurring in this way, since the syphoned sample showed no *Str. agalactiæ*. Attention was therefore directed to the treatment of the sore. All teats were smeared four times daily for 21 days with penicillin cream containing 5,000 units per gram, using 10 grams of cream daily. The cream was also inserted for a few millimetres into the LH teat canal. Seven days after the commencement of treatment, *Str. agalactiæ* could no longer be detected in milk samples taken in the ordinary way, whilst the healed sore failed to yield cultures from the eleventh day onwards. Swabs and milk samples examined 25 times in the subsequent four months remained negative.

The above observations show that milk samples may become infected from an infected teat canal sore.

(b) The Treatment of Sores on Teats, Using CTAB Cream

Nine out of 24 cows (37.5 per cent) kept in a shed had persistent sores on their teats, in spite of treatment with penicillin cream, 1,000 units per gram, twice daily after milking over a period of two months. *Str. agalactiæ* was recovered from the sores of five of the nine cows (55 per cent) when examined on the 2.6.48. One month later, 3 per cent CTAB cream in lanette wax, lanoline and paraffin base was applied to the teat sores twice daily after milking. Five days after the commencement of the treatment the sores on the teats of four cows had healed; eight days after the commencement of the treatment the sores on the teats of two more cows had healed; and on the 14.6.48, i.e., eleven

days after the commencement of the treatment, the sores on the teats of two of the remaining three cows had also healed. Thus, during a period of eleven days, using 3 per cent CTAB cream, the sores on the teats of eight out of nine cows were healed and *Str. agalactiæ* could no longer be recovered. In spite of this treatment, one cow (No. 988) still had an unhealed sore, from which *Str. agalactiæ* was recovered on the 11.6.48, but not on the 14.6.48.

No skin irritation or lesion was observed on the teats treated with 3 per cent CTAB cream either during the eleven days' treatment or up to three days after the last application.

(c) The Treatment of Sores on Teats, Using CTAB and Penicillin

Farm A. This herd consisted of 31 cows, seven of which (23 per cent) had sores on their teats. When examined on 23.6.48, the sores on six of these seven cows were infected with *Str. agalactiæ*. Treatment of the sores with 3 per cent CTAB cream, twice daily after milking, commenced on 23.6.48, and was continued up to 27.7.48, when the sores on four of the seven cows (57 per cent) were cured. From the unhealed sores on the three remaining cows *Str. agalactiæ* was recovered. From 27.7.48 to 16.8.48, the teats and udders of all cows were washed with 0.2 per cent CTAB aqueous solution, followed by water, twice daily after milking, and, when dry, penicillin cream (1,000 units per gram) was applied to the teats. When examined on 16.8.48, one cow had a healing sore on the left hind teat, and swabs taken from it and from all remaining teats were negative.

Farm B. Examination of this herd on the 29.6.48 showed that ten out of 26 cows (38 per cent) had *Str. agalactiæ*-infected sores on their teats. Treatment commenced on 30.6.48, with 3 per cent CTAB cream applied to the teat sores twice daily after each milking. Clinical and bacteriological examination carried out on the 21.7.48 showed that seven of the ten cows (70 per cent) were cured, and swabs taken from the teats of all cows were negative. Further treatment from 22.7.48 up to 9.8.48 consisted of washing the teats and udders of all cows with 0.2 per cent CTAB aqueous solution, followed by water, twice daily after milking, and, when dry, penicillin cream (1,000 units per gram) was applied. Examination carried out on 9.8.48 showed the teats to be quite free of sores or lesions, but *Str. agalactiæ* was still recovered from the teats of two cows.

It seems, therefore, that (1) the dead tissue in unhealed teat-sores, smeared twice daily with milk during wet milking or stripping, is a medium specially suited for the multiplication of *Str. agalactiæ*, and (2) CTAB and penicillin cream can encourage healing in a large proportion of the sores which may be a permanent and potential source of infection to the udder-secreting tissue and to the milk samples taken from the non-infected quarters.

Summary and Conclusions

The finding that *Str. agalactiæ* can survive up to three weeks on various objects in an infected cowshed, up to 26 days on some parts of the skin, and that it can multiply and persist in sores on the teats of non-infected udders

indicates that infection causing mastitis may arise from a source other than inside the bovine udder. The significance of the presence of *Str. agalactiæ*-infected sores on teats and at the outlet of the teat canal has been pointed out as a potential source of infection to the udder-secreting tissue and to the milk samples taken from non-infected udders. Efforts were directed to find the best method of eliminating *Str. agalactiæ* from these sources.

The effect of various disinfectants at different concentrations on *Str. agalactiæ* was tested *in vitro* and *in vivo*, to find the best agent which would remove or reduce to a minimum the number of organisms (a) from the skin, bearing in mind the existence of natural skin auto-disinfection, and (b) from contaminated objects and materials usually found in the environment of infected herds.

To test the efficiency of the various disinfectants, special selective liquid and solid media were used. Sterile milk was chosen as the medium in some of this work, as milk is an agent in the spread of infection in the herd and because the effect of disinfectants on *Str. agalactiæ* in the presence of the organic matter in milk is in some respects similar to the conditions in which these disinfectants act on the skin and on the contaminated objects outside the udder, where the organic matter present protects the organism and so reduces the power of disinfecting agents.

The results of the experiments showed that CTAB in aqueous solution and in cream, and iodine solution were the most satisfactory disinfectants for the skin, while CTAB aqueous solution and formaldehyde gave the best results for the cowshed and its fittings. Of the disinfectants tested, 0.1 to 1 per cent CTAB aqueous solution proved the most satisfactory for dairy utensils.. CTAB and penicillin creams appeared to be efficient in the treatment of sores on the teats.

The minimum effective concentration of CTAB for the disinfection of the skin by spraying or brushing was 0.5 to 1 per cent solution. Three per cent CTAB cream in lanoline, lanette wax and paraffin base was useful for teat disinfection and for the treatment of teat sores, provided that the skin reaction and sensitivity were observed and treatment discontinued when necessary. Penicillin cream 1,000 to 5,000 units per gram used alone, or following washing with 0.2 per cent CTAB aqueous solution and boiled water, was useful for the treatment of some persistent sores on the teats. Two per cent Chloros was used for the disinfection of the skin, care being taken to ensure that the available chlorine had not deteriorated and that spraying or brushing was carefully carried out, but was found to be much less effective than CTAB. The disinfection of a loose-box and its fittings, e.g., the wall, window ledge, cleaning utensils, milking stools, etc., by spraying with CTAB 0.2 per cent aqueous solution was effective. Formaldehyde vapour allowed to act for 24 hours on the *Str. agalactiæ* normally found on the objects in an infected cowshed—e.g., cleaning utensils, milking stools, sacks, clothing of cowshed attendants, etc.—was also very effective as a bactericidal agent. The use of this method of disinfection could be recommended for infected equipment on farms, providing that suitable accommodation for the purposes exists and that proper safety precautions are taken.

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One per cent CTAB aqueous solution can be used with safety without causing irritation to the skin or to the mucous membrane of the eye, while 0.1 to 0.2 per cent is satisfactory for the routine washing of the udder before milking. Three per cent CTAB cream can safely be used on the skin of the teats, 1 per cent cream on the udder skin, and two per cent cream on the remaining parts of the skin. When using CTAB, observations should be made, as some individual cows may show particular sensitivity of the skin to this agent.

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CONTROL AND PREVENTION OF NEUROLYMPHOMATOSIS (Fowl Paralysis)*

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WHILE a general understanding of the ætiology, symptomatology, morbid anatomy and micro-pathology of this important disease is both interesting and useful to all concerned in the poultry industry, a minute consideration is limited essentially to the pathologist and research worker.

Control and prevention, however, are aspects which will appeal most to the practical poultry farmer. It is readily admitted that our knowledge of the disease is yet incomplete, but as a result of experimentation certain facts have been elicited, a knowledge of which has permitted the institution of practical measures for successfully combating the disease.

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What are the facts on which these measures depend? Briefly, the disease is due to a virus which may be present in the droppings of affected birds or in the egg. Apparently healthy adult birds may be "carriers." Young birds are most susceptible; the period of greatest susceptibility is probably during the first four weeks of life. Birds over eight weeks are more resistant to infection. There is considerable difference in resistance and susceptibility; these are qualities which are inherited and can be enhanced by selection.

Much of this knowledge is the result of research in America, but a considerable amount of work has been done in Scotland and I propose to concentrate on it, firstly because I am naturally more familiar with it, and secondly because much of it was carried out under environmental conditions comparable to those pertaining on any well-run poultry farm. I also consider that we had two very valuable additional advantages denied American workers: the work was begun soon after the disease made its appearance in this country and a pedigree stock of known history was available for control purposes (the flock of Brown Leghorns maintained by Dr. Greenwood at Edinburgh University).

Hygienic Measures, including Isolation

It soon became evident that, to be successful, isolation must commence immediately after hatching, and all contact with adult stock, direct or indirect, should be avoided for at least six to eight weeks. The degree of isolation possible under practical conditions will depend on the type of farm and the facilities and staff available, and will be governed to an important extent by the severity of the disease problem, e.g., a high incidence in older stock would call for more stringent measures and the maximum degree of protection of chicks from infection, particularly during the earlier part of the critical period. The efficacy of isolation was proved experimentally by Johnston and Wilson (1937 and 1939) by rearing two large groups of chicks, the produce of the same parent stock, from day old, under widely differing environmental conditions: (a) on completely fresh ground and in thoroughly sterilised houses; and (b) on land where serious losses had occurred in successive years. The experiment lasted for two years and at the end of that time mortality due to fowl paralysis in stock on the clean ground was 1.11 per cent, compared with 31.4 per cent in the birds on the contaminated ground. Fuller details are shown in Table 1.

TABLE 1

	Parent Colony	Fresh Ground
Number of chicks (day old)	458	1,015
Number of pullets (2 months)	207	450
Total mortality per cent	76.8	20.9
Fowl paralysis „ „	31.4	1.11
Average weight at 6 months	3.24 lb.	3.87 lb.
Egg production per pullet	75.1	217.3

In another experiment where isolation commenced when chicks were eight weeks old there was no material reduction in the incidence of the disease.

The disease has been repeatedly produced in chicks at Eskgrove by placing them on infected land at an early age; and in every case controls, kept elsewhere on clean ground or intensively, have remained healthy (Wilson, 1944). On at least three occasions the disease has been diagnosed in pullets from an egg-laying test and in sister pullets from the home flock almost simultaneously; this is further confirmation that subsequent happenings are associated with early environmental conditions rather than with those obtaining at the crucial period when symptoms appear. The question of direct egg transmission is confusing and experimental results are conflicting. There is considerable evidence that it does occur and that the disease may be spread to susceptible chicks in the incubator. I am inclined to believe, however, that this is not such an important factor in causing serious losses as the introduction of infection during the first few weeks of life. All our work supports this contention, and it may well be argued that the occurrence of a small number of cases in isolation is due not to direct egg transmission but to incomplete isolation. It is of interest that cases never occurred in our special isolation units prior to the appearance of coccidia; we do not associate this parasite as a causal or even as a predisposing factor, but merely as a criterion of the breakdown of complete isolation.

The possibility that the virus may sometimes be present on the outside of the egg shell as a result of fæcal contamination must be considered; routine fumigation of eggs soon after setting, as practised in the control of Salmonellosis, will eliminate this as a potential source of infection.

There will perhaps be some who do not believe in isolation, even partial, as, of course, it must always be even under the best practical working conditions, and consider that it is better to get chicks outside on grass as soon as possible in order to produce the " hardy " chick even at the expense of loss of a proportion of them. There may be some basis for this in dealing with a disease where recovery from a sub-lethal infection produces immunity (e.g., coccidiosis), but this argument is unsound in neurolymphomatosis, where resistance depends on factors unaffected by environment. There are others to whom the intensive system is anathema, but we have shown experimentally under controlled conditions that where land is badly contaminated the incidence of the disease is greatly reduced by recourse to this system. This is supported by the behaviour of the disease in Dr. Greenwood's flock since its appearance in 1937. As is well known, no culling is carried out: yet the incidence throughout the years has remained negligible. This is not due to any inherent immunity in the chicks, as it has been shown that they develop the disease readily when exposed to infection.

Development of Resistant Lines

It has already been noted that certain birds appear to be resistant to the disease while others are susceptible. It must be clearly understood that such resistance does not depend in any way on prior exposure to the disease, but on the genetic constitution of the individuals concerned. Many stocks have been considered resistant but have speedily succumbed when exposed to infection; the appearance of the disease therefore depends on the level of infection to which

a flock is exposed, in conjunction with the degree of inherited resistance or susceptibility it possesses. There is a further factor which modifies the expression of the disease—the development of age resistance. If exposure to heavy infection is delayed, losses in a susceptible stock may be slight or even absent. Since exposure to infection during the critical period is the only criterion of resistance, to build up a resistant strain would entail the retention of affected birds on the breeding farm and the deliberate exposure of chicks to infection—a policy few breeders would contemplate. Without this exposure, freedom from disease, associated with absence of infection, might wrongly be interpreted as an expression of resistance and would result in the further production of susceptible stock.

It has been suggested that this type of programme might be carried out by State-aided breeding stations with the object of producing large numbers of male birds possessing a high degree of resistance for distribution to the industry. The many difficulties such a project would entail are at once apparent. A modified programme based on these principles can be carried out by the breeder who has the disease in his stock, by breeding from the survivors of an outbreak after careful and rigorous selection. There must always be a modicum of risk in this procedure, as amongst the survivors there may be carriers of the disease, but it seems preferable to the slaughter policy often practised in earlier years when the disease made its appearance on a farm. This was followed by the introduction of fresh stock from a supposedly (though often mistakenly) free flock, which almost invariably resulted in a fresh outbreak.

Much has been written of the value of eye colour as an indicator, but the shape of the pupil and its response to light stimuli are more important than colour. The golden-red iris is a highly prized selling factor, and birds possessing it will continue to be selected for breeding purposes—and rightly so—but it is no indication or measure of resistance to fowl paralysis.

If the disease position is serious pullet breeding should not be practised, for although fowl paralysis can occur in three- and four-year-old birds and even older, the highest incidence is in the pullet year. It is probable that even in older birds, infection takes place early in life; the virus remains viable and, as a result of lowered resistance for some reason or another, the state of equilibrium is ended and disease develops. The owner of a " closed " flock is, of course, in a strong position and should be able to keep the disease in check.

Diagnostic Tests

Research on fowl paralysis has been hampered by many difficulties, the two most important being: first, the absence of a method of diagnosing the disease in the apparently healthy bird (i.e., before symptoms are shown or in the " carrier " in which symptoms may never appear); and, secondly, the difficulty in reproducing experimentally the disease at will. Tests such as the agglutination test for *S. pullorum* or the tuberculin test are of great value, but, notwithstanding much work, a reliable test for the diagnosis of carriers of fowl paralysis is not yet available. Kissling (1947) described a leukoagglutination

test carried out much in the same way as the rapid whole-blood test—by mixing on a plate a small quantity of serum and an antigen made up of a suspension of lymphocytes, stained with methylene blue. We have repeated his work and have also prepared modified antigens, but difficulty has been experienced in interpreting the results of the test. There is often a very narrow dividing line between what is apparently a positive result, characterised by clumping accompanied by clearing, and the granular reaction which is assumed to be without pathological significance. We are not yet in a position to carry out this test, but it is a line of work which is being continued; for if some similar relatively simple procedure could be successfully evolved it would be of great value to the industry.

Therapeutics

From time to time accounts of the successful results of the administration of various medicaments have been recorded. I do not propose to deal with this aspect, as I consider that attempted curative treatment is contraindicated. Birds showing symptoms should always be destroyed. They represent susceptible stock which should not be perpetuated and are a potential reservoir of infection.

Summary

Some aspects of the control and prevention have been briefly discussed. The position can be summed up appropriately by a paragraph taken from an earlier paper which is still apposite:—

"In the light of our present knowledge it would appear that control and prevention may best be achieved by a careful selection of the breeding pen, discarding members whose progeny have shown evidence of the disease but returning pen sisters whose offspring have remained healthy, together with a strict isolation of young stock during the first few weeks of life: the use of fresh ground for rearing purposes or where this is impracticable recourse to the intensive system during the critical period."

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DIGEST CULTURE MEDIA USING PAW-PAW (CARICA PAPAYA) IN THE TROPICS

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THE paw-paw, fruit of the tree *Carica papaya,* is known to secrete a strong enzyme — papain — capable of digesting meat protein. The purified product is marketed in the form of a white powder. *Carica papaya* grows in the tropics and is abundant over most of West Africa, though it does not appear to be deliberately cultivated to any great extent. The crude commercial product is exported from the West Indies and East Africa.

The enzyme is powerful and rapid in its action on meat protein, and has been used in the preparation of bacteriological culture media. Asheshov (1941) outlines methods of preparing papain digest media, and notes that the enzyme is active at a relatively high temperature, and at *pH* values around neutrality. Pearston (1944), citing Asheshov, gives details for routine papain digest media preparation using Papainum (B.D.H.). The use of the enzyme does not, nevertheless, seem to be at all widespread in laboratories.

In Sierra Leone the paw-paw tree is very common, and it occurred to the writer that the abundant supply of fruit could be used to produce a suitable digest medium in the new laboratory of the Veterinary Department, set up in early 1949. At that time literature and precise information regarding the site of the enzyme in the plant, the optimum concentration, temperature, *pH*, etc., was not immediately available, and some time was spent in determining these factors. Results obtained were confirmed by the literature references eventually obtained.

Various parts of the plant were tried, including the juice of the ripe fruit, the pulp of ripe and unripe fruit—which were both relatively inactive—and the milky exudate which oozes from the cut surfaces of stem, leaf, and unripe fruit—which is active in varying degrees. The best source is the milky exudate from unripe fruit. The skin of the fruit is lightly pierced or scored with a sharp knife in a number of places and the exudate collected; the flow is small and ceases rapidly, a coagulum forming on the wounds which is scraped off and harvested also. The exudate flows from immediately beneath the fruit skin; if the pulp is deeply pierced the exudate appears to seep back into it and be lost. Very young fruits bleed well but the quantity is small; the ripe fruit which has turned yellow does not bleed. Insufficient flow is obtained from leaf or stem to be worth while. The same fruits have been bled repeatedly at intervals with no apparent loss of potency in successive batches, but the fruit is eventually spoiled.

The exudate was tested for digestive activity fresh in liquid form, and dried and ground to a powder. The fresh material is difficult to handle and coagulates easily, particularly in contact with water. The powder has been obtained here by collecting the exudate in shallow enamel trays, and solidifying rapidly in the sun, then drying overnight in the incubator at 37° C. The solid

material is then scraped off the trays, powdered in a mortar, and bottled. Samples kept in bottles at room temperature during the hottest season showed no loss of potency after 43 days.

The procedure adopted for the preliminary work was to use 10 grammes of minced beef, previously freed from fat and gristle, and suspend this in 30 or 40 c.c. of water in two-ounce round bottles plugged with cotton-wool. The exudate was added as a proportion of the weight of minced meat. The bottles were held at different temperatures for varying periods of time, and results assessed on the appearance, on titration of 10 c.c. of supernatant fluid or filtrate to *pH* 7.0 in a comparator using N/10 sodium hydroxide, and on the coagulation occurring when the neutralised supernatant or filtrate was boiled. Control bottles containing no paw-paw exudate were always included for comparison. The appearance after digestion was found to give a good indication of potency: the meat is reduced to a small amount of brownish sludge which contains a greater or lesser amount of undigested meat particles, and the supernatant fluid is coloured pale yellow. Chloroform was added to the bottles at first, but as it was found that good digestion proceeded quickly at temperatures approaching 60° C. this was abandoned.

Concentrations of 1 per cent, 2 per cent and 3 per cent were tried initially, and later serial dilutions of 0.09 per cent to 1.5 per cent were tested, also concentrations up to 12 per cent. The activity falls noticeably at 0.75 per cent, but no advantage could be found using concentrations greater than 2 per cent. The concentration finally adopted in bulk was 1.3 per cent. Trials at *pH* 8.4, and with the addition of hydrochloric acid to 1 per cent, both inactivated the enzyme. The prior adjustment of the meat/water mixtures to *pH* 7.0 with 30 per cent sodium carbonate was not found to be of any advantage. Digestion was noted to proceed very rapidly in the first 30 minutes, and in bottles to which enzyme had been added prior to raising the bottles to 100° C. in the steamer appreciable digestion had taken place during the hotting-up process before the enzyme was inactivated by steaming. This inactivation was confirmed by steaming an enzyme/water mixture, cooling, adding the requisite quantity of meat, and testing for digestion, which fails to occur.

The necessity for further temperature tests disappeared when a copy of Pearston's article came to hand, in which a temperature of 65° C. is recommended. Temperatures around 60° C. had already proved useful, and a standard temperature of 65° C. maintained for 3½ hours was adopted.

Considerable difficulty had been experienced in successfully "wetting" the powdered exudate, and a preliminary rubbing-up with a little water in a test-tube, using a glass rod, had been tried. Pearston mentions the use of a little glycerol, and this was found satisfactory.

Full-scale preparation of batches of media was successfully tried out, using the following materials and procedure:—

Minced beef, 1½ lb.
Water, 3 litres
Dried powdered paw-paw exudate, 10 grammes
Sodium chloride, 15 grammes

Rub up the dried powdered paw-paw exudate in a mortar with 40 to 50 drops of glycerol, and wash this into the meat/water mixture. Bring to 65° C., and hold for 3½ hours with occasional stirring. Boil for 2 or 3 minutes, allow to settle for 5 minutes, and strain through coarse cloth. Check the volume, replace loss, and add the sodium chloride. Adjust to *pH* 7.6. Boil or steam to precipitate, and filter. At this stage the medium is normally distributed and sterilised in the autoclave and stored. Agar is added for solid medium.

Where, however, as in Sierra Leone, the production of contagious bovine pleuro-pneumonia culture vaccine requires the preparation of large batches of serum broth, the medium is filtered through a coarse paper rapidly after *pH* adjustment, cooled, 10 per cent ox serum added, and the whole sterilised by filtration through Ford's Sterimat SB. It is then distributed aseptically.

Both procedures have been tried, also the production of agar medium, all successfully.

The main test organism has been the causal organism of bovine contagious pleuro-pneumonia, which is very critical in its growth requirements. Growth repeatedly obtained in the paw-paw digest serum broth described, and using a vaccine strain of this organism of about 90 generations, was equal to that obtained in the 2 per cent peptone meat infusion serum broth in normal use, and superior to Wright's broth with serum. It will be appreciated that peptone is both expensive and in short supply at the present time. It should also be noted that digest media using pig stomach are in successful use in some laboratories for the production of contagious bovine pleuro-pneumonia culture vaccine.

Owing to the fact that the laboratory in which this work was done was not fully equipped or in proper operation, a variety of test organisms was not readily available. A number of adventitious contaminants were tried, however, consisting of micrococci, and gram-negative and gram-positive bacilli, and all gave copious growth. *Brucella abortus,* strain S.19, also gave good growth.

Having regard to the recommendations by Asheshov and Pearston that the digest can be usefully diluted before use, some of the broth prepared here as described was diluted to half strength and put up both as solid and liquid medium. A single trial with the *Br. abortus* S.19 appeared to give good results with both full-strength and half-strength agar medium, but the half-strength broth with 10 per cent ox serum, and sterilised by filtration, failed to support full growth of the organism of contagious bovine pleuro-pneumonia comparable with that obtained in the routine peptone infusion medium, on the single occasion when this was tried.

Attempts were made to purify the crude material obtained from the paw-paw fruits, to concentrate the enzyme. The fresh exudate forms a thick gelatinous precipitate with water. The effect of organic solvents was tried, and alcohol was found to give a powdery suspension with both the fresh and the dried material, which is much easier to handle. At the same time some of the exudate appeared to go into solution. Attempts were made to extract

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the exudate, using different concentrations of alcohol; physiological saline; and 30 per cent glycerin; extracting at 60° to 65° C. for 2 to 3 hours. After extraction the material was divided roughly into soluble and insoluble portions, and tested separately for digestive activity. Saline and 30 per cent glycerol gave no fractionating effect and were discarded. Alcohol at 30 per cent was found to give a fractionating effect to some extent, and enzyme being apparently soluble. It was noted that the effect of treating a filtrate of the 30 per cent alcohol extraction with excess of absolute alcohol was to give a white precipitate which was freely soluble in water. This was accordingly filtered off, dissolved in water, and tested for digestive activity. No sharp fractionation could, however, be determined, the activity being distributed in decreasing amount through the various fractions. The procedure was therefore abandoned as being too cumbersome for ordinary use and offering no great advantage over the crude dried powder.

Contagious bovine pleuro-pneumonia vaccine prepared using the paw-paw digest medium has not been tried in the field, but a single test on a few cattle caused no harmful or unusual reaction.

No further work was carried out as the writer became due for leave in England.

Summary

A successful attempt to devise a relatively simple procedure for the production of digest media using locally available papain in the form of a dried powdered exudate from the fruit of *Carica papaya* in Sierra Leone is described.

It is suggested that such a medium, after suitable trial, might be useful for the production of contagious bovine pleuro-pneumonia culture vaccine, as well as for other purposes.

Unsuccessful attempts to concentrate the enzyme by simple purification of the exudate are described.

Acknowledgments

The approval of the Director of Veterinary Services, Sierra Leone/Gambia, has been given for the publication of this material. Thanks are also due to Dr. Reid at the Government Laboratory, Freetown, and to Mr. R. J. Bromfield in London, both of whom supplied references on request.

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SOME STRANGE CASE HISTORIES IN AN EGYPTIAN VETERINARY PRACTICE; Illustrating the Need for Providing Adequate Clinical Services to the Animal-owning Public

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THE parctising veterinary surgeon is often called to attend a case when all attempts on the part of the owner himself, and often of a multitude of neighbours, to deal with it successfully have failed. The veterinary surgeon has often found to his cost that his own skill was to be tested against the traditional practices of the neighbourhood, which, in themselves, would form a most fertile field for investigation by the mythologist. Above all, the veterinarian needs to maintain his composure in such circumstances. The late Professor Macqueen, at the beginning of his lectures on veterinary obstetrics, used to give vivid description of the scene which often confronted the veterinarian after he was called out, usually late at night, to a cowshed in a lonely country district. After depicting that scene, Macqueen's counsel was to say nothing but proceed calmly to examine the animal itself carefully; after that, the veterinarian was to wash his hands, take out his pipe and sit down quietly to think out the best procedure to adopt to bring away the calf.

In Egypt the conditions as so described are not rare. Indeed, the above would be considered a very mild occurrence.

Notes on a Few Incidents

CASE NO. 1.

A cow-buffalo. According to the owner, who was a poor and ignorant peasant, the animal had been a first-class milking buffalo but suddenly, for no apparent reason, the milk yield had diminished, the appetite had become depraved, and the animal had changed completely in its behaviour. Some local remedies were given, but the symptoms became aggravated so much that at last he decided to call for veterinary advice.

When I saw the case, the animal was depressed, dull and sluggish, and had completely lost her appetite. There was no fever, the mucous membranes were slightly pallid. No precise history concerning the case could be elicited, and the owner hotley denied having performed any manipulative treatment. I began treatment with rectal examination, in the first place withdrawing the fæcal contents, when my hand encountered a solid spherical body, the nature of which could not be determined as it was in a state of partial decay. Proceeding, another such body was found which was also much decayed, and then still a third, a fourth and a fifth and, on breaking the last, it could be distinguished as a raw onion. Confronted with

these objects the owner still denied having introduced them, but, after withdrawing several more, and speaking to him with some asperity, he confessed the truth which was that, when he had noticed the cow-buffalo had failed to improve in condition after administering some of the usual local remedies, a neighbour had advised him to insert raw peeled onions into the rectum, one by one, until she got well. When I asked him what was the number he had inserted, he replied that he could not remember. In any event, the number of intact ones which I removed was over a dozen. The animal afterwards was treated with enemata, given an oily drench and kept under my personal supervision. It recovered completely in a week.

CASE NO. 2.

A cart-horse. When brought to me for treatment it was suffering from an intense conjunctivitis and inflammation of the cornea of the right eye. The eyeball appeared to be in a very serious condition as if it had suffered from some severe accident or injury. On asking the attendant what had happened, he unhesitatingly and frankly confessed that he had found the horse in the morning running a watery discharge at the eye: the advice of a neighbour had been to sprinkle pepper into it. As there was no improvement after the sprinkling but, on the contrary, the eye trouble got much worse, and the animal was in much pain, he decided to call for veterinary advice which, fortunately, was within a few hours after the first symptoms were noticed. After proper treatment, the eye recovered without complications.

CASE NO. 3.

A donkey, eight months old. It was suffering from hip-lameness. The owner, a poor and very stupid man, had been advised by someone to seton the hip, and had carried out the advice with such thoroughness that he had inserted three setons instead of one. The animal was in great pain, with an enormous inflammatory swelling in the region of the hip. The case was admitted to the hospital of the Veterinary School within three days of the operation and the setons were removed. The donkey recovered and was discharged after two months.

CASE NO. 4.

A foal, newly-born. The owner had been advised that to strengthen its limbs and maintain them in good alignment he ought to tie each limb tightly with a piece of cord, as with a tourniquet, above the knees and hocks. This had resulted in such considerable swelling below the cords that he thought he had better obtain veterinary advice. After removing the cords, the swellings gradually disappeared, and the foal grew up into a fine animal.

CASE NO. 5.

A camel. Admitted into the hopsital of the Veterinary School with a seton inserted just above one eyelid and suffering from a marked inflammatory reaction of the eye. When questioned, the owner declared that he had always

been under the impression that setoning was beneficial for inflammatory conditions of the limbs and some other parts of the body, and so it might also be of benefit to an inflamed eye. After removal of the seton above the eye and proper treatment, considerable improvement took place, though complete resolution did not take place.

CASE No. 6.

A country-bred horse, used for drawing a light cart. As the animal had been off food for some time and was debilitated, the owner sought local advice, which was to fire the breast to improve the appetite. The result was that the horse developed tetanus, which ended fatally.

CASE No. 7.

Cart-horse. Had shown symptoms of fever. The owner was advised to bleed the horse from the ears by slitting the concha from its tip. The fever did not improve and the ears became badly infected. The owner decided then to call for veterinary assistance. After prolonged treatment, the animal recovered.

CASE No. 8.

A mare, four years old. A good-looking, handsome animal which had developed sprained tendons in both forelegs and had been successfully treated by me with the usual remedies. My subsequent advice was to rest the mare for a suitable period before returning her to work. It transpired, however, that the owner was persuaded by a friend, contrary to the advice I had given him, to fire the tendons and, not content with firing in the usual way, had proceeded to carry out that operation so vigorously as to sever the tendons of both forelegs. On returning to me for veterinary advice, all I could do was to destroy it immediately.

CASE No. 9.

A donkey. It had shown symptoms of colic. The owner had been advised to give a local colic drench which was administered through the nostrils instead of through the mouth. This practice is not uncommon in Egypt. The donkey rapidly developed pneumonia. Veterinary treatment, when sought, was of no avail, and the animal died on the same day.

CASE No. 10.

A horse. Reported to have been badly bitten on the middle of the back by another horse. The owner had treated the case himself with some local remedies, with the result that the wounds became badly infected. Extension of infection took place to the deeper tissues, causing marked necrosis of the spinous processes of the dorsal vertebræ. After about six weeks, the back presented such an advanced gangrenous appearance that the owner decided to seek veterinary advice. When I saw the case there was a highly offensive odour which could be detected a long distance away. Portions of the dorsal vertebræ could be detached easily by hand. For humanitarian reasons I ordered the destruction of the horse.

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A recital of cases such as given certainly makes sordid reading. If an apology is required for submission of such experiences for publication, it is that they represent, indeed, I believe, a fair sample of what comes to the notice of a qualified veterinarian in Egypt, who has made some attempt during his career to supply the animal-owning public of that country with a kind of professional aid which, at the moment, they sorely need but which is almost completely lacking. As is well known, nearly all the graduates from the Veterinary School seek and obtain employment immediately in Government service, mainly to discharge certain veterinary police and clerical functions, which rarely gives them any opportunity to develop a professional ability and be able to render immediately veterinary aid to the individual animal. It is true that the immense majority of animal owners are unable to pay much in the way of professional fees. It is contended, however, that a modern State must confess itself bankrupt in the matter of providing its people with adequate social services if it cannot devise ways and means of safeguarding animals and their owners against ignorance and victimisation of the sort that has been delineated above. The animal-loving section of the public now forms by far the largest proportion of the population of Egypt, and animal-ownership, whether of a horse, donkey, working bullock or a buffalo, etc., is very often the sole form of private possession allowed them in the social economy. The veterinarian, who for some reason or other has entered into private practice, and come into direct personal contact with animals and their owners, sees many tragedies, of which the above are a few selections taken at random. Something positive must be done by those in authority to cleanse our country of these inhuman practices which are the product of ignorance and neglect.

Acknowledgment

Some help in writing these records was received from J. T. Edwards, visiting Professor and temporary Dean of this Faculty.

THE STOMACH OF THE CAMEL

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Introduction

THE stomach of most domesticated animals has been subjected to detailed description, but very little has been recorded about that of the camel.

The ability of camels to live without water for a long period, and the fact that some owners, engaged in illicit drug traffic, have been able to secrete metal containers in the so-called "water sacs" within the stomach, led the writer to make this anatomical study.

Literature

Chauveau (1891) considers the reticulum to be a part of the rumen which is distinguished by the presence of deep cells. He notes the narrow opening existing between the omasum and the abomasum. He adds that the omasum shows deep areolæ, instead of leaves.

Lesbre (1903) states that the reticulum is ovoid, and that it communicates with the abomasum by an opening measuring from 8-10 cm. in circumference. He mentions the absence of the omasum, and regards the part of the stomach of the camel connecting the reticulum with the duodenum to be a single reservoir, which he calls the abomasum.

Referring to the water sacs, Lesbre describes the presence of two mucous membrane pillars surrounding two groups of aquatic sacs, which he considers as dilatations of the wall of the rumen. He also adds that some of these water sacs, consisting of about 50 primary aquatic cells, are also arranged in transverse rows numbering 15.

Leese (1927) states that the water sacs are formed of a number of cells arranged in rows at right-angles to each other. Each water cell opens in the cavity of the rumen by a square mouth. The muscle bundles around the cells act as sphincters to retain their contents, while contraction of the walls and floor causes evacuation of these contents. He also notes the connection between the rumen and the reticulum by a large opening, inside which and across the small curvature of the reticulum runs the œsophageal groove. He also refers to the ill-defined demarcation between the omasum and the abomasum.

Sisson (1938) describes the stomach of the horse as a large dilatation of the alimentary canal between the œsophagus and the small intestine, its capacity being not more than 2-4 gallons. It is a sharply curved, U-shaped sac situated in the dorsal part of the abdominal cavity behind the diaphragm and liver, mainly to the left of the median plane. He refers to its two curvatures, and notes that the lesser is very short, dorsal in direction, and extends from the œsophagus to the pylorus. With reference to the extremities, he mentions that the left extremity is rounded, forming a cul-de-sac—the saccus cœcus—while the right (pyloric) is much smaller. With regard to the œsophagus, he mentions

that it joins the stomach very obliquely, and that its opening is closed by the sphincter cardiæ and numerous folds of mucous membrane.

The œsophageal orifice—the cardia—is situated at the left end of the lesser curvature, about 20-25 cm. from the left extremity. The pylorus is the opening into the intestine; its position is indicated externally by a distinct constriction, while internally it presents a circular ridge, caused by a ring of muscular tissue (the sphincter pylori).

The description of the stomach of the horse by Chauveau (1891), M'Fadyean (1922), Strangeways (1919), and Bradley (1923) is similar to that of Sisson.

The stomach in ruminants is very large, according to Chauveau, being divided into four separate pouches. It fills the greater part of the abdominal cavity, with a capacity of not less than 55 gallons. He mentions that the rumen, in which the œsophagus opens, constitutes nine-tenths of the total mass, while the other three (reticulum, omasum and abomasum) form a short chain, continuous with the left and anterior portion of the rumen. He considered the abomasum to be the true stomach, and the other three compartments represent, like the left sac in solipeds, œsophageal dilatations. With regard to the stomach of the ox, the same author mentions that the left sac of the rumen surpasses the right at its two extremities; on the other hand, in the case of the sheep and the goat he adds that the right sac is longer than the left. He mentions that the omasum is larger than the reticulum in the ox, and that in the sheep and the goat it is smaller.

The description of the stomach of the ox by Sisson is similar to that of Chauveau, except that the former adds that the œsophagus opens into the stomach on a sort of dome—the *atrium ventriculi*—and is continued through the reticulum by the œsophageal groove, and that from the ventral end of the latter a groove transverses the ventral wall of the omasum, thus giving a direct path from the reticulum to the abomasum for finely divided food or fluid. The same author gives the capacity of the stomach of the ox to be 25-60 gallons, with great variation, depending on age and size of the animal. He concludes that the omasum is smaller and nearly rounded, and that its cavity is occupied to a considerable extent by about a hundred longitudinal folds (laminæ omasi), separated from each other by a series of folds, decreasing gradually in height till it forms a series of very low folds or lines, while the cavity of the rumen is covered with a brown mucous membrane, which is studded with large papillæ (1 cm. long).

The same author describes the stomach of the pig as a large single sac of a capacity of 1-2 gallons, with a flattened, conical, blind pouch—the *diverticulum ventriculi*. Chauveau adds that it is simple, like the horse, but less curved, and that the cardia lies nearer to the left extremity. He also refers to the greater width of the œsophageal opening, and to the prolongation of its mucous membrane around the cardia. The same author describes the stomach of both dog and cat as pear-shaped, with a dilated cardia, which is nearer to

the left extremity of the organ than in other animals. He also refers to the remarkably regular and undulated folds of its mucous membrane when the stomach is empty. Again according to Sisson, the stomach of the dog is relatively large, capacity 6-7 pints, and its lesser curvature is nearly straight and vertical.

Materials and Method

The stomachs of about 50 camels were examined carefully in the abattoirs of both Cairo and Belbeis in both their normal position and outside the carcass. Other camels were subjected to detailed dissection in the anatomy department of the Faculty after injecting them with the usual preservative solution..

Macroscopical Examination

It was observed that the stomach of the camel, like that of other ruminants, is composed of four compartments—rumen, reticulum, omasum and abomasum —marked off externally from each other by constrictions, which are very faint between omasum and abomasum.

Examination of the interior of the rumen of the camel reveals that its greater part is lined by a smooth, pale mucous membrane which is loosely attached to the submucosa by a large amount of areolar tissue. In the empty state of this organ the mucous membrane is thrown into numerous folds, which have no definite arrangement and which become entirely obliterated when the organ is distended with food material.

The remaining portion of the rumen, though, is lined by similar mucous membrane, and is interrupted by a number of well-defined muscular bands enclosing between them what is known as water sacs. These water sacs form three separate groups, the largest of which is situated in front and to the right, while the smallest group is in the apex of the rumen. The largest water sac is marked off from the rumen by a thick, muscular pillar, which corresponds with a well-defined groove externally separating the apex of the rumen from the region of the water sacs.

On the right muscle fibres from the muscular layer of the rumen converge to form the before-mentioned muscular pillar, which proceeds across the wall of the rumen to the left side, forming a slight arch, the convexity of which is anterior and terminating in the œsophageal region by dividing into ventral and dorsal muscular bands which enclose the œsophageal opening. From the latter point the dorsal band is continued across the dorsal surface of the rumen to end abruptly after a short course of about 6-7 inches. On the other hand, the ventral band gradually merges into the muscular layer of the ventral surface of the rumen.

The largest group of these water sacs is divided into seven compartments by means of parallel muscular bands which spring from the anterior convex border of the muscular pillar already mentioned. These muscular bands proceed anteriorly across the floor and continue for a short distance on its roof, where they gradually merge into the general muscular wall of the rumen. Each

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of the compartments of this group of water sacs is in turn partitioned transversely into aquatic cells, varying in number from 3-8, by folds of mucous membrane, which enclose between their layers thin, yellow elastic bands at the orifies of the aquatic cells. These aquatic cells vary in size, depth and arrangement. In the floor of the water sac the aquatic cells are wide, deep, and simple. These features are marked in the aquatic cells which are situated immediately in front of the muscular pillar. On the other hand, the aquatic cells on the sides and roof of the water sac are small, less deep, and partially subdivided at their bases.

The second water sac is found occupying the posterior part of the floor of the rumen. In structure it is similar to the previously described water sac and it differs only in the direction of the muscular bands, which are transversely disposed. These bands are six in number and enclose between them the aquatic cells, which are also simple, resembling those present in the floor of the previous water sac.

The third, and the smallest, of all the groups of water sacs is located in the left side of the apex of the rumen. This consists of about 5-7 small and shallow aquatic cells of the simple type.

It was noticed that all the above-mentioned water sacs are wider at their bases than at the orifices, and that the largest one never attains a depth of more than 10 cm., and its capacity is approximately 200-300 c.c.

The œsophageal groove extends from the œsophageal opening along the roof of the rumen to the rumino-reticular opening, where it transverses the dorsal part and then continues along the lesser curvature of the reticulum, to terminate at the reticulo-omasal opening. In the first part of its course this groove proceeds in a forward direction to the reticulo-ruminal opening. From the latter point it turns to the left across the lesser curvature of the reticulum to its termination. The œsophageal groove measures about 12 inches long and 2 inches wide. It is bounded by two mucous ridges, the right of which is more prominent than the left all along its course. On the other hand, the left ridge is well defined in the posterior half of its course only. The mucous membrane lining the œsophageal groove is somewhat paler in colour, thinner, and is more intimately attached to the submucous layer than that lining the rumen itself. It should be noted that the muscular layer of the rumen is well developed in the floor, and particularly so in the region of the muscular pillar.

Microscopical Examination

Sections through the water sacs of a fresh stomach reveal the presence of some minute, separate glands, which do not differ in structure from those observed along different parts of the digestive tube of other domesticated animals.

Conclusion

Many theories have been advanced by former workers on the function of the water sacs.

Leese (1927) stated that it is thought that water can be stored in these sacs and brought up into the mouth cavity whenever the camel wants to quench its thirst, and he concludes that the function of these water sacs is the moistening of their contents by their active secretion.

Lesbre (1903) states, according to some authors (Phere and Evered Home), that these water sacs function as a store for water for future use, and that the muscular bands surrounding the orifices of the water sacs can close them to the extent of excluding the entrance of solid alimentary matter. He also refers to other author's opinion that the aquatic cells not only store water taken, but also produce watery secretion. Lesbre, however, expressed his disagreement with these authors, because of the absence of glandular tissue on microscopical examination which could account for their statement. He also explains the presence of the green coloration of the contents of the water cells that was collected by Good and Trustworthy to the entrance of some of the contents of the rumen in the relaxed water sacs after death.

All my observations on this matter support the opinion of those who state that the water sacs function as a store for water. Camels have the habit of drinking large quantities of water before setting out on a long journey during which they may not obtain any more. Moreover, many travellers have saved their own lives by killing camels in order to quench their thirst by the water they knew to be found in these water sacs.

The presence of the greenish water previously noted by Good and Trustworthy was noted in all post-mortems under examination. The cause of this greenish coloration was adequately explained by these authors.

Acknowledgment

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CORRESPONDENCE

ECOLE VETERINAIRE DE LYON

BRITISH veterinary surgeons have a deep regard for the ancient School of Veterinary Science at Lyons, not only on account of its age and great tradition, but also because we are indebted to Lyons for St. Bel, the founder of the London School, and, incidentally, of modern veterinary science as it is known amongst all the English-speaking peoples.

We have received from our friends at Lyons, and through the agency of Professor P. Goret, a letter, addressed to their British colleagues, extending a cordial invitation to attend a congress in Lyons, to be held during the month of June, 1950.

It gives us great pleasure to publish this letter, and the accompanying programme of events.

We are sure that those of our colleagues who can make the trip to Lyons will be assured of a warm welcome; they will enjoy their sojourn in the ancient city and gain much profit from the scientific discussions.

To the Editor, "British Veterinary Journal."

I write to ask if you will be willing to insert in your next issue a provisional programme of a congress of veterinary surgeons to be held at Lyons. The papers and discussions all deal with the subject, "Sterility in Cattle." We will be happy if a number of our English colleagues will attend and take part in the discussions. I will be able to send soon the final programme to those colleagues who wish to attend. In the meantime I enclose the provisional programme.

In the hope that you will be able to publish this letter and programme, I send you, Mr. Editor, my best wishes.

P. GORET, *Professor.*

[EDITOR.—We would suggest that those colleagues who would like to attend this congress should communicate their intention to Professor Goret.]

VETERINARY CONGRESS OF THE SCHOOL OF LYONS

June 23, 24 and 25, 1950.

PROVISIONAL PROGRAMME

FRIDAY, 23

Morning—

Opening of the Congress.

Professor Jung: "General Physiology of the Genital Apparatus of the Bovine Animal."

Professor Bethelon: "Ætiological Diagnosis of the Sterility of the Bovine Animal."

Afternoon—

Dr. Vicard: "Sterility in the Bovine Animal: The French Practitioner's Point of View."

Dr. Pellaton: "Sterility in the Bovine Animal: The Swiss Practitioner's Point of View."
Discussion.

SATURDAY, 24

Morning—
Professor Brion: "General Therapeutics of the Sterility of the Bovine Animal."
Dr. X: "Comparative Pathology: Sterility in Women."
Afternoon—
Syndical meeting—Meeting of the Order.
Syndical and Professional Outlooks.
Dr. Labattut: "Collective Prophylaxy of the Sterility of the Bovine Animal."
Discussion.
Closing of the Congress.

SUNDAY, 25

Morning—
Meeting of the Association of Ex-Students and Friends of the School of Lyons.
Meeting of the Lyons Veterinary Sciences Society on the subject of meat inspection.
Lecturers: Professor Lucam and Dr. Fontenaille.
Noon—Banquet.
Afternoon—Dancing and recreation.
Evening—Annual Show by the Students, followed by a Ball.

REVIEWS

ANIMAL DISEASES IN SOUTH AFRICA, by M. W. Henning. Second Edition. London: Gordon & Gotch, Ltd., 65s. od. net.

THE title of this work might well be "Diseases of Animals in Africa," or even "Diseases of Animals in the Tropics and Sub-tropics." To the uninitiated the title might be taken to mean that all the diseases included in Professor Henning's book occurred in South Africa. Of course, it is possible that any of them might be introduced, but for South Africa's sake we hope that a disease such as rinderpest, which was banished from the Union in 1903, will never return. It is important, however, that veterinary surgeons working in South Africa should be well versed in any disease which might occur, for as recently as 1942 rinderpest actually threatened the Rhodesias and the Union. In this respect this book is very complete and adequately covers any disease of animals likely to be met with in that part of the world.

A modern textbook on tropical veterinary medicine has been urgently required for some time, and now the second edition of "Animal Diseases in

South Africa" almost fills this gap. Since the first edition, published in 1932, the tempo of veterinary research has greatly increased, and there is a mass of new information on the various diseases of domestic animals which has been published in different parts of the world. Professor Henning is to be congratulated on collecting and collating this information, very much up to date, in a most efficient and readable manner. The section devoted to diseases caused by plant poisons in the earlier edition has been left out of this volume, as this subject has been dealt with in a separate work by Steyn, so that the extra information incorporated in this edition has not made the format unwieldy.

In tropical veterinary medicine there are so many different sciences involved, such as entomology, plant ecology, botany, and so on, that with certain subjects it is difficult to give an adequate overall picture and yet keep within reasonable limits. Professor Henning has overcome this very well, and the chapters on trypanosomiasis, rabies, and the tick-borne diseases are not only up to date, but are most excellent monographs of the subjects under review.

The book is arranged in three sections dealing with bacterial diseases, protozoal diseases, and diseases caused by viruses and rickettsias. The chapters are well set out and the discussion on the historical aspects of each disease makes most interesting reading and whets the literary appetite for the more technical subject matter which follows. General interest is also added by numerous references to similar diseases or the same disease as it occurs in human beings. A very complete bibliography is given at the end of each chapter.

South Africa has led the way in the control of a number of diseases not necessarily confined to that country, and the anthrax spore vaccine originally elaborated at Onderspoort is in wide use in Great Britain and elsewhere, whilst the work on mineral deficiencies and its relation to botulism, under the name of lamsiekte, are classical achievements in veterinary science. The description of these conditions in Henning's book are excellent.

"Animal Diseases in South Africa" can be confidently recommended to all veterinarians and veterinary students, and it will be invaluable to those working in tropical and sub-tropical countries.

Perhaps Professor Henning might be persuaded to round off his work by including a few tropical diseases of Asiatic origin, and entitling it "Animal Diseases in South Africa and the Tropics."

EUGEN FROHNER LEHRBUCH DER TOXIKOLOGIE FUR TIERARZTE, by R. Volker (Direktor des Pharmakolog, Instituts der Tierärztl, Hochscule, Hannover). Sixth edition, 1950. Pp. xx + 404. Price DM 35.60. Stuttgart: Ferdinand Enke.

FOR many years Fröhner's text-book on Veterinary Toxicology was regarded as the standard German work on this subject, but it is now twenty-two years since the last edition made its appearance. A new edition, prepared by Professor Völker, is therefore to be welcomed, for the previous edition has long been out of date.

In order to include as much as possible of the new material which has been accumulating, the book has been entirely revised and rearranged. After an introductory section on general toxicology, the individual substances are dealt with under the headings of inorganic compounds, organic compounds, poisonous fungi and plants, and animal poisons. The general plan has been to give a brief account of the chemical compound, followed by a description of the conditions under which the substance is most likely to give rise to poisoning—this heading is termed " ætiology "—and then an account of the pathogenesis, symptoms, post-mortem findings and treatment. Owing to the need for limiting the size of the book, the chemical detection of the various substances has not been included. This detracts somewhat from the value of the book, for although chemical toxicology is admittedly a subject for the qualified analyst, especially if legal proceedings are contemplated, nevertheless there are many occasions on which the knowledge of how to carry out simple tests, such as the Reinish test, may be of the greatest assistance in confirming a case of poisoning without having to resort to a detailed analysis.

On the other hand, the number of substances listed is perhaps greater than is necessary in a work of this nature. Here the approach seems to have been more that of the pharmacologist than the toxicologist since such drugs as the sulphonamides, and even adrenaline, are included. Many of the commercial solvents are also mentioned.

The publishers have produced a book of excellent quality, and there is no doubt that it will be found to be a most useful addition to veterinary literature.

NOTICES

New Features of Milward's "Vim" Syringes

THE growing use of autoclaving and hot air sterilising has made tough, hard-wearing syringes essential.

In the past, hot air sterilising made imperative the all-glass syringe, which, however, proved vulnerable in the nozzle. One authority estimated that chipped nozzles accounted for half their total breakages.

The glass and metal syringe is, of course, more sturdy, and Messrs. Midward's, whilst retaining the advantages of the individually mated glass plungers and barrels, have attacked and overcome the jointing problem by developing a cement capable of withstanding a temperature greatly in excess of the 160 degrees C. prescribed by the Medical Research Council in War Memorandum No. 15. This provides an ample margin of safety even in the most extreme temperatures.

Another important aspect of the syringe service is the permanence of the syringe markings. Even engraved markings are hard to see unsupported by colour, and Milward's again have met this problem by the introduction of a heat treatment, or vitrifying process, whereby the marking pigment becomes an

XUM

integral part of the glass itself. Even if a large part of the glass is ground away the markings still remain clear and distinct.

A further essential feature claimed is the good fit between plunger and barrel. Milward's use a new and unique method of grinding, evolved from lessons learned over many years in this country and abroad, to give these components a fit of unprecedented accuracy.

MOBILE HOT WATER

We recently had the opportunity of witnessing a demonstration of Calor Gas Water Heaters, marketed by Ranalah, Ltd., Burbage, Leics.

These were seen working on a farm where there was no electricity or town gas, but, when coupled to a water main, hot water was supplied instantly at the economical rate of approximately three gallons for 1d.

These appliances fill a long-felt want on the isolated farm, both for washing down cows, cleaning milking utensils, and a hundred and one other uses. They will also provide hot water in the kitchen and bathrooms of the homestead. From the veterinary point of view they can be used to great advantage in country consulting and operating rooms, as well as kennels, stables, etc., and greatly simplify the question of hygiene in all its aspects.

They are so portable that they can be taken out into a field and used where there is a stream, tap, storage tank, or pails of water.

Weather has no effect on them; they will even work perfectly in a full gale. To veterinary surgeons these heaters will prove a boon. Really hot water anywhere in a few seconds.

The makers will be pleased to send you further particulars by return of post.

HEPAMINO BROCHURE.—Evans Medical Supplies, Ltd., have issued a new brochure entitled "Hope Still Remains," dealing with Hepamino, the original Proteolysed Whole Liver.

The illustration on the cover is "Pandora," by Harry Bates.

Publishers' Notices

The British Veterinary Journal, with which is incorporated The Veterinary Journal, is published monthly and copy for advertisements should be in the hands of the publishers—Baillière, Tindall and Cox—not later than the 20th of the preceding month, if proof is required. Tel.: Temple Bar 3386.

Letters for the Journal, literary contributions, reports, notices, books for review, exchanges, new instruments or materials, and all matter for publication (except advertisements) should be addressed to the Editor.

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